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Prostate cancer risk and polymorphism in 17 hydroxylase (CYP17) and steroid reductase (SRD5A2)

Ruth M.Lunn, Douglas A.Bell, James L.Mohler[2] and Jack A.Taylor[1,3]

Laboratory of Computational Biology and Risk Assessment and [1]Epidemiology Branch and Laboratory of Molecular Carcinogenesis, National Institute of Environmental Health Sciences, Research Triangle Park, NC 27709 and [2]Department of Surgery, Division of Urology, University of North Carolina, Chapel Hill, NC, USA

[3]To whom correspondence should be addressed at Molecular and Genetic Epidemiology Section, NIEHS, Mail Drop A3–05, PO Box 12233, Research Triangle Park, NC 27709, USA
Email: taylor@niehs.nih.gov

Prostate cancer is the most common malignancy in males and is the second most common cause of cancer mortality in American men. Polymorphisms have been identified in two genes, the 17-hydroxylase cytochrome P450 gene (*CYP17*) and the steroid 5-reductase type II gene (*SRD5A2*) that are involved with androgen biosynthesis and metabolism. The *CYP17* A2 allele contains a T→C transition in the 5′ promoter region that creates an additional Sp1-type (CCACC box) promoter site. The *SRD5A2* valine to leucine (V89L) polymorphism has been correlated with lower dihydroxytestosterone levels. We tested genotypes in 108 prostate cases and 167 controls along with samples (*n* = 340) from several different ethnic groups. The *CYP17* A2 allele (combined A1/A2 and A2/A2 genotypes) occurred at a higher frequency in Caucasian patients with prostate cancer (70%) than in Caucasian clinical control urology patients (57%), suggesting that the A2 allele may convey increased risk for prostate cancer [odds ratio (OR) = 1.7, 95% confidence interval (CI) = 1.0–3.0]. Blacks and Caucasians had a similar frequency of the A2 genotype (16 and 17%, respectively) while Taiwanese had the highest frequency (27%). The *SRD5A2* leucine genotype was most frequent in Taiwanese (28%), intermediate in Caucasians (8.5%) and lowest in Blacks (2.5%). Genotypes having a *SRD5A2* leucine allele were somewhat more common in prostate cancer cases (56%) than in controls (49%) (OR = 1.4, 95% CI = 0.8–2.2) but this difference was not significant. These results support the hypothesis that some allelic variants of genes involved in androgen biosynthesis and metabolism may be associated with prostate cancer risk.

Introduction

Prostate cancer is the most common malignancy and is the second most common cause of cancer mortality in American men (1). Despite its impact on public health, little is known about its etiology. Studies of risk factors such as occupation, diet, smoking, alcohol and sexual activity are inconclusive (2–5). However, age, ethnicity and family history clearly affect the risk of prostate cancer (6–8). Eighty percent of prostate cancer develops in men over 65 years of age. Prostate cancer incidence is 20- to 30-fold higher in American Blacks than in Asians and is intermediate in American Caucasians (6,7). Men having two or more affected first-degree relatives are 5 to 11 times more likely to develop prostate cancer (9).

Steroid hormones appear to be important determinants of prostate cancer risk. Androgens are potent prostate mitogens (10) and elevated androgen levels may be associated with prostate cancer risk. Gann *et al.* (11) reported increasing risk of prostate cancer with increasing levels of plasma testosterone; however, other studies have failed to observe an association (12–14) and plasma levels of testosterone may not accurately measure the amount of free testosterone in the prostate (11). Testosterone is synthesized from cholesterol by a series of enzymatic reactions involving several of the cytochrome P450 enzymes (for a review, see ref. 15) (Figure 1). Of interest is CYP17, which catalyzes two sequential reactions in the steroid biosynthesis pathway. The first step is the conversion of pregnenolone to 17-hydroxypregnenolone, which is subsequently converted to C19 steroid dehydroepiandrosterone, a compound with androgenic activity (15). A T→C transition (A2 allele) in the 5′ promotor region of *CYP17* creates an additional Sp1-type (CCACC box) promoter site, suggesting that the A2 allele may have an increased rate of transcription. Although the effect of this allele on phenotype has not been demonstrated, it has been associated with elevated serum estrogen and progesterone concentration in nulliparous women (16).

Testosterone is converted to 5α-dihydroxytestosterone (DHT) in the prostate by the enzyme 5α-reductase, type II (SRD5A2). DHT binds to the androgen receptor (AR) forming a complex (AR–DHT) which translocates to the nucleus and transactivates target genes (17,18; Figure 1). A decrease in the length of the polymorphic trinucleotide CAG repeat in the *AR* gene enhances transactivation (19). Shorter repeats are found in Blacks and are associated with increased prostate cancer risk (17,20).

SRD5A2 activity (conversion of testosterone to DHT) varies in different ethnic populations, and polymorphisms in the *SRD5A2* gene have been identified which correlate with enzyme activity and/or ethnicity (21–24). Ross *et al.* (25) reported higher SRD5A2 activity in Blacks than in Asians, which parallels observed ethnic differences in prostate cancer risk. A valine to leucine polymorphism at codon 89 (V89L) has been reported, with the leucine allele (L) associated with lower steroid 5α-reductase activity and a lower prevalence in Blacks (21).

Using a case control study design, we studied the association between genetic polymorphism and prostate cancer risk in the putative protective allele (leucine) of the *SRD5A2* gene and the hypothesized risk allele (A2) of the *CYP17* gene.

Abbreviations: AR, androgen receptor; AR–DHT, androgen receptor–dihydroxytestosterone complex; BPH, benign prostate hypertrophy; CI, confidence interval; CYP17, 17-hydroxylase cytochrome P450; DHT, dihydroxytestosterone; OR, odds ratio; PCR, polymerase chain reaction; SRD5A2, steroid 5-reductase type II.

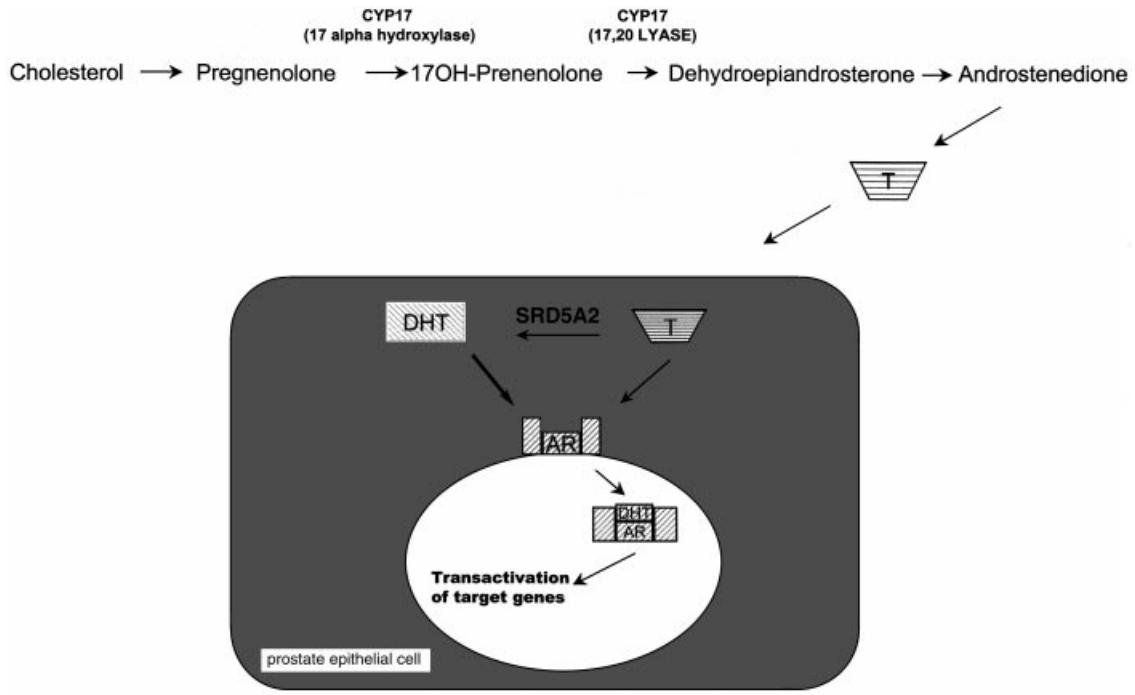

Fig. 1. Model of steroid-dependent prostate carcinogenesis. A simplified diagram of the biosynthesis pathway of testosterone and its conversion to DHT is shown. Testosterone is converted into DHT in the prostate epithelial cells. Both DHT and testosterone can bind the AR; DHT binds the AR with 4- to 5-fold higher affinity than testosterone. The activated AR–DHT complex binds to androgen-responsive elements of target genes involved in prostate cell proliferation. T, testosterone (36).

Materials and methods

Subjects

Subjects have been described previously in a study of vitamin D receptor polymorphisms and prostate cancer risk (26). Briefly, they consisted of 108 consecutive prostatectomy patients (96 Caucasians and 12 Blacks) at the University of North Carolina Hospitals in Chapel Hill, NC and 167 male non-cancer controls (159 Caucasians and eight Blacks) enrolled at the urology clinics at University of North Carolina Hospitals and at nearby Duke University Medical Center in Durham, NC. Most control subjects presented to the urology clinic for evaluation and treatment of voiding symptoms due to prostatic hypertrophy or impotence. Control subjects had no history of cancer other than non-melanoma skin cancer. Blood was collected from both cases and controls and DNA was extracted using standard phenol–chloroform methodology.

As an independent measure of allele frequency in different ethnic populations in North Carolina, gene frequencies were also determined for North Carolinian Black and Caucasian populations who have been described previously and been used in other genotyping studies (27–30). Briefly, DNA was extracted from blood samples drawn from a community-based sample comprised of over 400 healthy unrelated Blacks and Caucasians from Durham and Chapel Hill, NC. A subset ($n > 100$) of each ethnic group (Blacks and Caucasians) were used for estimating *CYP17* and *SRD5A2* genotype frequencies. A larger sample of 176 Caucasians was used for *SRD5A2* genotyping in order to provide a more precise estimate of the uncommon LL genotype. Allele frequency was also measured in 110 Taiwanese. Placenta DNA (kindly provided by Dr L.L.Hsieh) was isolated from 110 unrelated full-term maternity patients with uncomplicated pregnancy at the Chang Gung Memorial Hospital, Taiwan (31).

Genotyping

CYP17. Polymorphisms in *CYP17* were detected using polymerase chain reaction (PCR)–restriction fragment length polymorphism analysis. A 629 kb DNA fragment including the A1/A2 polymorphic site was amplified from 50 ng genomic DNA using the PCR primers *CYP17214F* (TCCTGAGCCCAG-ATACCAT) and *CYP823R* (CCGCCCAGAGAAGTCCT). The PCR reaction consisted of an initial 4 min denaturation step followed by 30 cycles of 30 s at 94°C, 30 s at 60°C and 30 s at 72°C. The presence of the A2 allele creates a *Msp*AI restriction site in the PCR fragment. The PCR product was digested with *Msp*A1 for 2 h at 37°C and run on 3% Nusieve 3:1 agarose gels (FMC Bioproducts, Rockland, ME). The A1/A1, A1/A2 and A2/A2 genotypes resulted in 577; 577, 305 and 272; and 305 and 272 bp digestion products, respectively. A 52 bp fragment was present in all samples due to an invariant *Msp*A1 site that served as an internal control for complete digestion.

SRD5A2. Fifty nanograms of genomic DNA was used in a 15 µl PCR reaction containing PCR primers *SRD5A2415F* (TCCAGAAGTTGCCGCATCAG) and *SRD5A2039R* (CGGTGCGCGCTCCAC) and 1× additive Q reagent (Qiagen, Santa Chatsworth, CA). Q reagent provided higher yields in optimization experiments. Cycling conditions were the same as for *CYP17* except the annealing was at 65°C. The presence of the valine (V) at base 1007 results in an *Rsa*I restriction site. The 639 bp PCR product encompassing the polymorphic site was digested with *Rsa*I for 2 h at 37°C and the digestion products were separated using 2.5% Metaphor agarose gels (FMC Bio-products). Polymorphic specific digestion products consisted of 100 and 20; 120, 100 and 20; and 120 bp fragments for the VV, VL and LL genotypes, respectively. Two fragments, 404 and 168 bp, were present in all samples due to invariant *Rsa*I sites and served as internal controls for completed digestion.

Statistical analysis

Differences in genotype or allele frequency between the various ethnic groups (Blacks, Caucasians and Asians) were tested by pair-wise comparisons of χ^2 2×3 or 2×2, contingency table analysis. The association between genetic polymorphisms and prostate cancer was evaluated by traditional 2×2 table analysis. Fisher exact odds ratio (OR) and the corresponding 95% confidence intervals (CIs) were calculated for each ethnic strata, and the Mantel–Haenszel weighted ORs (OR_{MH}) were calculated for combined strata using the software EGRET [Epidemiological Graphics, Estimation, and Testing package analysis module (PECAN), v.0.22].

Results

Ethnic variation

We estimated genotype frequencies of *CYP17* A1/A2 and *SRD5A2* V/L genes in sample populations of North Carolinian Blacks and Caucasians and in Taiwanese (Table I). The genotype distribution for both *CYP17* and *SRD5A2* were found in Hardy–Weinberg equilibrium for each ethnic group. While the frequency of the *CYP17* A2 allele (and resulting genotypes) was similar in Blacks (0.36) and Caucasians (0.38), the A2 allele was significantly more prevalent in Taiwanese (0.52), with $P < 0.01$ and $P = 0.04$ for Taiwanese versus Blacks

Table I. *SRD5A2* and *CYP17* genotype and allele frequency among Taiwanese and North Carolinian populations of Caucasians and Blacks (community-based sample)

	NC community population		
	Blacks[a]	Caucasians[a]	Taiwanese[a]
CYP17	*n* = 115	*n* = 115	*n* = 110
A1/A1	51 (44%)	47 (41%)	26 (24%)
A1/A2	46 (40%)	48 (42%)	54 (49%)
A2/A2	18 (16%)[bc]	20 (17%) [bd]	30 (27%) [cd]
Allele frequency (A2)	0.36	0.38	0.52
SRD5A2	*n* = 118	*n* = 176	*n* = 108
VV	77 (65%)	73 (41%)	16 (15%)
VL	38 (32%)	88 (50%)	62 (57%)
LL	3 (2.5%)[ef]	15 (8.5%)[eg]	30 (28%)[fg]
Allele frequency (L)	0.19	0.38	0.56

[a]Allelic distribution of *CYP17* and *SRD5A2* genotypes in Blacks (B), Caucasians (C) and Taiwanese (T) sample populations were in Hardy–Weinberg equilibrium.
[b]Genotype frequency for B versus C was not significantly different; $\chi^2 = 0.311$, $P = 0.856$.
[c–g]Genotype frequency for the following were significantly different:
[c]B versus T, $\chi^2 = 11.7$, $P = 0.003$.
[d]C versus T, $\chi^2 = 8.29$, $P = 0.0159$.
[e]B versus C, $\chi^2 = 17.2$, $P = 0.002$.
[f]B versus T, $\chi^2 = 67.6$, $P < 0.001$.
[g]C versus T, $\chi^2 = 31.5$, $P < 0.001$.

Table II. CYP17 genotypes among prostate cancer patients and controls

	Cases	Controls	OR[a]	95% CI	*P* value
Caucasians	*n* = 96	*n* = 159			
A1/A1	29 (30%)	68 (43%)	1.0 (ref)		
A1/A2	54 (56%)	73 (46%)	1.7	1.0–3.2	0.07
A2/A2	13 (14%)	18 (11%)	1.7	0.7–4.2	0.27
A1/A2 + A2/A2	67 (70%)	91 (57%)	1.7	1.0–3.1	0.05
Blacks[b]	*n* = 12	*n* = 8			
A1/A1	4 (33%)	4 (50%)			
A1/A2	6 (50%)	3 (37.5%)			
A2/A2	2 (17%)	1 (12.5%)			
A1/A2 + A2/A2	8 (67%)	4 (50%)			
Combined[cd]	*n* = 108	*n* = 167			
A1/A1	33 (31%)	72 (43%)	1.0 (ref)		
A1/A2	60 (55%)	76 (46%)	1.7	1.0–3.1	0.05
A2/A2	15 (14%)	19 (11%)	1.7	0.7–4.1	0.21
A1/A2 + A2/A2	75 (69%)	95 (57%)	1.7	1.0–3.0	0.04

[a]Fishers exact OR.
[b]OR and 95% CI not calculated due to small sample size.
[c]Test for trend (A1/A1, A1/A2, A2/A2); $P = 0.08$.
[d]OR_{MH}.

and Caucasians, respectively. The distribution of the S*RD5A* genotypes was significantly different between all three populations, with the L allele occurring the highest in the Taiwanese (0.56), intermediate in Caucasians (0.38) and the lowest in Blacks (0.19) (Table I).

Case-control study

Caucasian prostate cancer cases more frequently carried the *CYP17* A2 allele (70%) than did clinic controls (57%; OR = 1.7, 95%CI = 1.0–3.1; $P = 0.05$; Table II). The same magnitude of effect was apparent when comparing the referent A1 homozygotes with heterozygotes or with homozygotes for the A2 allele (Table II). Small sample sizes precluded calculations of OR for Blacks alone, but Blacks were included as a weighted strata in a combined OR_{MH} of 1.7, 95% CI = 1.0–3.0; $P = 0.04$.

Table III. CYP17 genotypes in prostate cancer stratified by age in Blacks and Caucasians

	Cases	Controls	OR[a]	95% CI	*P*-value
⩽64 years	*n* = 61	*n* = 78			
A1/A1	14 (23%)	32 (41%)	1.0		
A1/A2 + A2/A2	47 (77%)	46 (59%)	2.3	1.0–4.8	0.03
>64 years	*n* = 47	*n* = 89			
A1/A1	19 (40%)	40 (45%)	1.0		
A1/A2 + A2/A2	28 (60%)	49 (55%)	1.2	0.6–2.6	0.68

[a]OR_{MH}.

Stratified analyses were performed according to age at diagnosis (Table III). The cases and clinic controls were grouped according to the mean age of the Caucasian clinic controls (64 years) which was similar to the mean age of cases (63 years). Among those who were ⩽64 years at diagnosis, *CYP17* A2-containing genotypes were significantly associated with prostate cancer (OR = 2.3, CI = 1.0–5.4; $P = 0.03$). Among older men, there was some evidence of increased risk from the A2 allele although this was not statistically significant. There was no evidence of an association between genotype and aggressiveness of tumor when comparing cases with Gleason grade <7 with those with Gleason grade ⩾7 ($\chi^2 = 0.165$, $P = 0.684$; data not shown). Stage data were not available for analysis.

We were concerned that the controls included men with a clinical diagnosis of benign prostate hypertrophy (BPH) which could potentially be associated with differences in steroid metabolism. Therefore, we stratified the controls into those with BPH and those without BPH and evaluated the *CYP17* genotype distribution in these populations. The OR estimate for the *CYP17* A2 allele was higher when prostate cancer cases were compared with controls with BPH (OR = 2.3, 95% CI = 1.1–4.5) than when compared with controls without BPH (OR = 1.4, 95% CI = 0.7–2.7). However, these two estimates do not differ statistically from each other, and *CYP17* genotype distribution of controls with BPH did not differ from controls without BPH ($\chi^2 = 1.8$, $P = 0.18$) or from Caucasian community population ($\chi^2 = 0.95$, $P = 0.33$). We also evaluated other diagnoses of the clinical controls, e.g. impotence, and found no relationship with genotype distribution.

Comparison of cases and controls for the *SRD5A2* L allele revealed an association in the opposite direction to that predicted for this putative protective allele (Caucasians: OR = 1.4, 95% CI = 0.8–2.4) although this finding was not statistically significant (Table IV). The small number of Blacks precluded any analysis. No association was observed after stratifying by age or BPH classification (data not shown).

Discussion

We evaluated the association between prostate cancer and polymorphisms in the *CYP17* and *SRD5A2* genes, two genes involved in the biosynthesis and metabolism of testosterone. Because testosterone and DHT are potent prostate mitogens, elevated levels in prostate tissue are hypothesized to play a role in unregulated prostate growth and tumorigenesis and have been shown to be associated with increased risk of

Table IV. *SRD5A2* genotypes among prostate cancer patients and controls

	Cases	Controls	OR[a]	95% CI	*P* value
Caucasians	*n* = 96	*n* = 148			
VV	42 (44%)	77 (52%)	1.0		
VL	47 (49%)	58 (39%)	1.5	0.8–2.6	0.17
LL	7 (7%)	13 (9%)	1.0	0.3–2.9	1.0
VL + LL	54 (56%)	71 (48%)	1.4	0.8–2.4	0.24
Blacks[b]	*n* = 12	*n* = 8			
VV	5 (42%)	2 (25%)			
VL	6 (50%)	5 (63%)			
LL[b]	1 (8%)	1 (12%)			
VL + LL	7 (58%)	6 (75%)			
Combined[cd]	*n* = 108	*n* = 156			
VV	47 (44%)	79 (51%)	1.0		
VL	53 (49%)	63 (40%)	1.4	0.8–2.4	0.24
LL	8 (7%)	14 (9%)	0.9	0.3–2.6	1.0
VL + LL	61 (56%)	77 (49%)	1.3	0.8–2.2	0.32

[a]Fishers exact OR.
[b]OR and 95% CI not calculated due to small sample size.
[c]Test for trend (VV, VL, LL) $P = 0.63$.
[d]OR_{MH}.

prostate cancer (10,11). We found some evidence that the putative high activity allele (A2) of the *CYP17* gene, which would be predicted to increase levels of testosterone, may be associated with prostate cancer (OR = 1.7, 95% CI = 1.0–3.0; $P = 0.04$).

The association between the A2 allele and prostate cancer was present in all men and stratifying by age revealed greater risk among men who developed disease at an earlier age (OR = 2.3, 95% CI = 1.0–5.4; $P = 0.03$). Testosterone levels decrease and sex hormone binding globin levels increase with increasing age (32). Thus, the proposed increased production of testosterone due to the A2 allele may be most important in early onset disease. Interestingly, the high-risk allele of the AR receptor is also primarily a risk factor in men under 60 years of age (17). We found no association between the *CYP17* A2 allele and Gleason grade. However, because our sample was restricted to men undergoing prostectomy and who may, therefore, have less aggressive disease, we cannot rule out the possibility that the A2 allele may be associated with disease severity.

The observed difference between prostate cancer cases and clinical controls is unlikely to be the result of biased population sampling. Allele frequency in controls is consistent among Caucasians. There was no significant difference in our estimate of the A2 allele frequency from the Caucasian clinical controls (0.34, $n = 158$) than that of the Caucasian community population (0.38, $n = 115$; $P = 0.38$; Table I). Moreover, both of these allele frequency measurements are similar to that observed in Caucasian women controls from North Carolina (0.36, $n = 379$) and London (0.36 $n = 47$) (33) and not significantly different to two recent reports of Caucasian women controls from New York (0.42, $n = 148$) and Maryland (0.42, $n = 113$) (34,35).

One concern of our study was that the inclusion of BPH patients in our clinical controls could potentially bias results since BPH may be related to steroid hormone levels. The association between the A2 allele and prostate cancer was greatest when the controls with BPH were used as a referent (OR = 2.3, 95% CI = 1.1–4.5). However, the distribution of C*YP17* A2 allele of Caucasian controls with BPH was not significantly different to that of Caucasian controls without BPH or the Caucasian community reference group, allowing us to use the combined BPH, non-BPH as the clinical control group

The A2 allele of the *CYP17* gene occurred at a higher frequency in Taiwanese than in Blacks or Caucasians, which does not correlate with ethnic differences in cancer incidence. Indirect correlation between allele frequency, ethnicity and cancer incidence can be misleading, and the association between *Cyp17* genotypes and prostate cancer remains to be tested in a Taiwanese population. The difference between risk due to genotype and risk due to ethnicity may be a result of the multiple genes involved in the predisposition to prostate cancer, or due to dietary or lifestyle differences between ethnic groups.

Following the hypothesis of Makridakis *et al*. (21) that the L allele of the *SRD5A2* gene should be protective for prostate cancer, we evaluated the genotype distribution in our case-control population. We did not detect evidence of a protective effect from the L allele in Caucasian men in our prostate case-control study and in fact found a slight, but not statistically significant elevated risk from the allele. The L allele is associated with lower serum levels of 3α-androstanediol glucuronide (AAG), which is a measure of 5α-reductase activity (21). Weak associations between serum AAG and prostate cancer have been observed (11). This is the first case-control study to our knowledge investigating the valine/leucine alleles of the *SRD5A2* gene. Studies of a TA repeat polymorphism in the 3′-untranslated region of the *SRD5A2* gene have also failed to demonstrate significant difference between cases and controls (18,23). Because the number of Blacks in our study was small, we were unable to evaluate the effect of the L allele in this population. The effect of various polymorphisms of the *SRD5A2* allele may vary in different ethnic groups, warranting more extensive multi-ethnic studies.

Although sex hormones appear to be involved in prostate carcinogenesis, the role of various polymorphisms in genes comprising the steroid biogenesis pathway are still unclear. Our study provides some evidence that the *CYP17* A2 allele may correlate with risk for developing prostate cancer, but does not support the hypothesis that the *SRD5A2* L allele is protective. Functional characterization of the different polymorphisms and their effect on steroid hormones in prostate tissue remain an important but relatively unexplored area.

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References

1. Landis,S.H., Murray,T., Bolden,S. and Wingo,P.A. (1998) Cancer statistics, 1998. *CA Cancer J. Clin*., **48**, 6–29.
2. Ilic,M., Vlajinac,H. and Marinkovic,J. (1996) Case-control study of risk factors for prostate cancer. *Br. J. Cancer*, **74**, 1682–1686.
3. Daviglus,M.L., Dyer,A.R., Persky,V., Chavez,N., Drum,M., Goldberg,J., Liu,K., Morris,D.K., Shekelle,R.B. and Stamler,J. (1996) Dietary beta-carotene, vitamin C, and risk of prostate cancer: results from the Western Electric Study [see comments]. *Epidemiology*, **7**, 472–477.
4. Lumey,L.H. (1996) Prostate cancer and smoking: a review of case-control and cohort studies [published erratum appears in *Prostate* (1996) **29**, 402]. *Prostate*, **29**, 249–260.
5. van der Gulden,J.W., Kolk,J.J. and Verbeek,A.L. (1992) Prostate cancer and work environment [see comments]. *J. Occup. Med*., **34**, 402–409.

6. Whittemore,A.S. (1994) Trends in prostate cancer incidence and mortality. In Doll,R., Fraumeni,J.F. and Muir,C.S. (eds), *Cancer Surveys*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
7. Parkin,D.M., Pisani,P. and Ferlay,J. (1993) Estimates of the worldwide incidence of eighteen major cancers in 1985. *Int. J. Cancer*, **54**, 594–606.
8. Neuhausen,S.L., Skolnick,M.H. and Cannon-Albright,L. (1997) Familial prostate cancer studies in Utah. *Br. J. Urol*., **79** (Suppl. 1), 15–20.
9. Steinberg,G.D., Carter,B.S., Beaty,T.H., Childs,B. and Walsh,P.C. (1990) Family history and the risk of prostate cancer. *Prostate*, **17**, 337–347.
10. Lee,C. (1997) Cellular interactions in prostate cancer. *Br. J. Urol*., **79** (Suppl. 1), 21–27.
11. Gann,P.H., Hennekens,C.H., Ma,J., Longcope,C. and Stampfer,M.J. (1996) Prospective study of sex hormone levels and risk of prostate cancer [see comments]. *J. Natl Cancer Inst*., **88**, 1118–1126.
12. Carter,H.B., Pearson,J.D., Metter,E.J., Chan,D.W., Andres,R., Fozard,J.L., Rosner,W. and Walsh,P.C. (1995) Longitudinal evaluation of serum androgen levels in men with and without prostate cancer. *Prostate*, **27**, 25–31.
13. Nomura,A.M., Stemmermann,G.N., Chyou,P.H., Henderson,B.E. and Stanczyk,F.Z. (1996) Serum androgens and prostate cancer. *Cancer Epidemiol. Biomarkers Prev.*, **5**, 621–625.
14. Vatten,L.J., Ursin,G., Ross,R.K., Stanczyk,F.Z., Lobo,R.A., Harvei,S. and Jellum,E. (1997) Androgens in serum and the risk of prostate cancer: a nested case-control study from the Janus serum bank in Norway. *Cancer Epidemiol. Biomarkers Prev.*, **6**, 967–969.
15. Waterman,M.R. and Keeney,D.S. (1992) Genes involved in androgen biosynthesis and the male phenotype. *Horm. Res*., **38**, 217–221.
16. Feigelson,H.S., Shames,L.S., Pike,M.C., Coetzee,G.A., Stanczyk,F.Z. and Henderson,B.E. (1998) Cytochrome P450c17alpha gene (*CYP17*) polymorphism is associated with serum estrogen and progesterone concentrations. *Cancer Res*., **58**, 585–587.
17. Stanford,J.L., Just,J.J., Gibbs,M., Wicklund,K.G., Neal,C.L., Blumenstein, B.A. and Ostrander,E.A. (1997) Polymorphic repeats in the androgen receptor gene: molecular markers of prostate cancer risk. *Cancer Res*., **57**, 1194–1198.
18. Feigelson,H.S., Ross,R.K., Yu,M.C., Coetzee,G.A., Reichardt,J.K. and Henderson,B.E. (1996) Genetic susceptibility to cancer from exogenous and endogenous exposures. *J. Cell Biochem*., **25** (suppl.), 15–22.
19. Choong,C.S., Kemppainen,J.A., Zhou,Z.X. and Wilson,E.M. (1996) Reduced androgen receptor gene expression with first exon CAG repeat expansion. *Mol. Endocrinol*., **10**, 1527–1535.
20. Ingles,S.A., Ross,R.K., Yu,M.C., Irvine,R.A., La Pera,G., Haile,R.W. and Coetzee,G.A. (1997) Association of prostate cancer risk with genetic polymorphisms in vitamin D receptor and androgen receptor [see comments]. *J. Natl Cancer Inst*., **89**, 166–170.
21. Makridakis,N., Ross,R.K., Pike,M.C., Chang,L., Stanczyk,F.Z., Kolonel, L.N., Shi,C.Y., Yu,M.C., Henderson,B.E. and Reichardt,J.K. (1997) A prevalent missense substitution that modulates activity of prostatic steroid 5α-reductase. *Cancer Res*., **57**, 1020–1022.
22. Reichardt,J.K., Makridakis,N., Henderson,B.E., Yu,M.C., Pike,M.C. and Ross,R.K. (1995) Genetic variability of the human SRD5A2 gene: implications for prostate cancer risk. *Cancer Res*., **55**, 3973–3975.
23. Kantoff,P.W., Febbo,P.G., Giovannucci,E., Krithivas,K., Dahl,D.M., Chang,G., Hennekens,C.H., Brown,M. and Stampfer,M.J. (1997) A polymorphism of the 5α-reductase gene and its association with prostate cancer: a case-control analysis. *Cancer Epidemiol. Biomarkers Prev.*, **6**, 189–192.
24. Davis,D.L. and Russell,D.W. (1993) Unusual length polymorphism in human steroid 5α-reductase type 2 gene (SRD5A2). *Hum. Mol. Genet*., **2**, 820.
25. Ross,R.K., Bernstein,L., Lobo,R.A., Shimizu,H., Stanczyk,F.Z., Pike,M.C. and Henderson,B.E. (1992) 5-alpha-reductase activity and risk of prostate cancer among Japanese and US white and black males. *Lancet*, **339**, 887–889.
26. Taylor,J.A., Hirvonen,A., Watson,M., Pittman,G., Mohler,J.L. and Bell, D.A. (1996) Association of prostate cancer with vitamin D receptor gene polymorphism. *Cancer Res*., **56**, 4108–4110.
27. Bell,D.A., Taylor,J.A., Butler,M.A., Stephens,E.A., Wiest,J., Brubaker, L.H., Kadlubar,F.F. and Lucier,G.W. (1993) Genotype/phenotype discordance for human arylamine *N*-acetyltransferase (NAT2) reveals a new slow-acetylator allele common in African-Americans. *Carcinogenesis*, **14**, 1689–1692.
28. Bell,D.A., Taylor,J.A., Paulson,D.F., Robertson,C.N., Mohler,J.L. and Lucier,G.W. (1993) Genetic risk and carcinogen exposure: a common inherited defect of the carcinogen-metabolism gene glutathione *S*-transferase M1 (GSTM1) that increases susceptibility to bladder cancer. *J. Natl Cancer Inst*., **85**, 1159–1164.
29. Stephens,E.A., Taylor,J.A., Kaplan,N., Yang,C.H., Hsieh,L.L., Lucier,G.W. and Bell,D.A. (1994) Ethnic variation in the CYP2E1 gene: polymorphism analysis of 695 African-Americans, European-Americans and Taiwanese. *Pharmacogenetics*, **4**, 185–192.
30. Watson,M.A., Stewart,R.K., Smith,G.B., Massey,T.E. and Bell,D.A. (1998) Human glutathione *S*-transferase P1 polymorphisms: relationship to lung tissue enzyme activity and population frequency distribution. *Carcinogenesis*, **19**, 275–280.
31. Hsieh,L.L. and Hsieh,T.T. (1993) Detection of aflatoxin B1–DNA adducts in human placenta and cord blood. *Cancer Res*., **53**, 1278–1280.
32. Wu,A.H., Whittemore,A.S., Kolonel,L.N., John,E.M., Gallagher,R.P., West,D.W., Hankin,J., Teh,C.Z., Dreon,D.M. and Paffenbarger,R.S.Jr (1995) Serum androgens and sex hormone-binding globulins in relation to lifestyle factors in older African-American, white, and Asian men in the United States and Canada. *Cancer Epidemiol. Biomarkers Prev.*, **4**, 735–741.
33. Gharani,N., Waterworth,D.M., Williamson,R. and Franks,S. (1996) 5′ polymorphism of the CYP17 gene is not associated with serum testosterone levels in women with polycystic ovaries [letter]. *J. Clin. Endocrinol. Metab*., **81**, 4174.
34. Helzlsouer,K.J., Huang,H.Y., Strickland,P.T., Hoffman,S., Alberg,A.J., Comstock,G.W. and Bell,D.A. (1998) Association between CYP17 polymorphisms and the development of breast cancer. *Cancer Epidemiol. Biomarkers Prev.*, **7**, 945–949.
35. Weston,A., Pan,C.F., Bleiweiss,I.J., Ksieski,H.B., Roy,N., Maloney,N. and Wolff,M.S. (1998) CYP17 genotype and breast cancer risk [In Process Citation]. *Cancer Epidemiol. Biomarkers Prev.*, **7**, 941–944.
36. Galbraith,S.M. and Duchesne,G.M. (1997) Androgens and prostate cancer: biology, pathology and hormonal therapy. *Eur. J. Cancer*, **33**, 545–554.

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